

This Page Is Inserted by IFW Operations
and is not a part of the Official Record

BEST AVAILABLE IMAGES

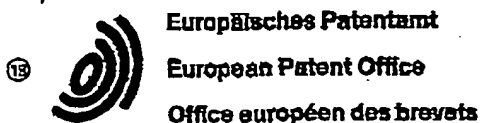
Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images may include (but are not limited to):

- BLACK BORDERS
- TEXT CUT OFF AT TOP, BOTTOM OR SIDES
- FADED TEXT
- ILLEGIBLE TEXT
- SKEWED/SLANTED IMAGES
- COLORED PHOTOS
- BLACK OR VERY BLACK AND WHITE DARK PHOTOS
- GRAY SCALE DOCUMENTS

IMAGES ARE BEST AVAILABLE COPY.

**As rescanning documents *will not* correct images,
please do not report the images to the
Image Problem Mailbox.**



(11) Publication number:

**0 115 627
A1**

(12)

EUROPEAN PATENT APPLICATION

(21) Application number: 83113070.3

(51) Int. Cl.: **A 61 K 37/02**

(22) Date of filing: 23.12.83

(30) Priority: 28.12.82 US 454128

(71) Applicant: Armour Pharmaceutical Company, 303 South Broadway, Tarrytown New York 10591 (US)

(43) Date of publication of application: 15.09.84
Bulletin 84/33(72) Inventor: Mufson, Daniel, 24 Hillside Drive, Thibault, N.Y. (US)
Inventor: Hanson, Musetta A., 1 Consulate Drive, Tuckahoe, N.Y. (US)(64) Designated Contracting States: AT BE CH DE FR GB IT
LI LU NL SE

(74) Representative: Patentanwälte Gröneck, Dr. Kinkeldey, Dr. Stockmair, Dr. Schumann, Jakob, Dr. Bazold, Melster, Hilgers, Dr. Meyer-Plath, Maximilianstrasse 58, D-8000 München 22 (DE)

(84) Enhancement of intranasal absorption of calcitonin by formulation with surfactants.

(57) A pharmaceutical composition for the treatment of disorders of bone metabolism which comprises an aqueous or non-aqueous medium suitable for intranasal administration and containing a therapeutically effective amount of calcitonin and a surface active agent.

EP 0 115 627 A1

4940

0115627

-7-

1 ENHANCEMENT OF INTRANSAAL ABSORPTION OF
 CALCITONIN BY FORMULATION WITH SURFACTANTS

5 The present invention relates to a novel method of
administering calcitonin to patients and to formulations
adapted for nasal administration.

10 Calcitonin is a polypeptide hormone isolated from
different organs in different species, including man and
salmon, or obtained via synthetic routes. Calcitonin is
15 recognized as being effective in diminishing hypercalcemia
and decreasing plasma phosphate concentrations in patients
with hyperparathyroidism, idiopathic hypercalcemia of
infancy, vitamin D intoxication, and osteolytic bone
metastases. While direct renal effects and actions on the
15 gastrointestinal tract are recognized, calcitonin is best
known for its effect on bone. Its use has proved to be
effective in diseases characterized by increased skeletal
resorption and abnormal bone formation, such as occurs for
example, in Paget's disease.

20 The method of administration of calcitonin is
predominantly by injection, although efforts were made in the
prior art to use other modes of administration, especially
for the treatment of localized conditions. While injectable
administration by physicians of calcitonin is proper for
25 short-term therapy, administration of calcitonin by injection
to patients in need of long-term calcitonin therapy has a
serious problem. Not only is it costly to patients to have
physicians do the administration of calcitonin for extended
periods of time but it is also painful and inconvenient. Nor

30

35

-2-

0115627

1 can calcitonin be given orally to patients as it will be
destroyed by the digestive juices in the gastrointestinal
tract.

5 In view of the foregoing, it is apparent that a
serious need exists for a different route of delivery of
calcitonin to patients suffering from conditions that require
prolonged calcitonin therapy.

Nasal preparations are known in the prior art.
Generally, nasal preparations comprise an oil-in-water or
10 water-in-oil emulsion or an oily solvent base suitable for
use on the mucous membranes, such as mineral or vegetable
oils and fatty acid esters and one or more chemicals which
are soluble in the base. Such preparations usually contain
one or more active drugs intended to alleviate or mitigate a
15 condition in the body by their adsorption into the blood
stream through the mucous membrane of the nose.

While small molecules such as propranolol are
efficiently absorbed intranasally, large molecules such as
calcitonin show little if any absorption. The purpose of
20 this invention is to find agents capable of increasing the
bioavailability of calcitonin so that cost of therapy is
reasonable. The prior art has also recognized that the nasal
absorption of certain drugs may be facilitated by the use of
surfactants in such nasal preparation. For example, insulin
25 and polypeptides were found to have an improved absorption
rate when used in a solution containing a surfactant.

30

35

-3-

0115627

1 It has now been found that hypercalcemia, Paget's
disease and other disorders of bone metabolism can be
advantageously treated by intranasal application of
calcitonin contain in a nasal preparation having an
5 absorption promoter and a buffer as essential ingredients.
Such preparations possess enhanced absorption across the
nasal mucosa when applied intranasally, but causes no
irritation or discomfort on extended use.

10 The present invention relates to a method for the
treatment of a mammal suffering from a disorder characterized
by high serum calcium which comprises intranasal application
of a nasal preparation containing a peptide having calcitonin
activity and an absorption promoting agent to effect control
of said disorders by transepithelial action.

15 According to the invention, calcitonin is
intranasally administered to a mammal via a novel dosage
form, such as a solution, ointment, or gel.

20 Calcitonin is a peptide hormone of 32 amino acids
with a disulfide bond at 1-7 in the amino terminus of the
molecule. These first seven amino acids with the disulfide
bond seem essential for activity and this sequence is
preserved from species to species. Calcitonin, as used
herein, means not only peptides having a structure
corresponding to one of the naturally occurring hormones, and
25 which may be naturally or synthetically produced, but also
related peptides having calcitonin activity.

The amount of calcitonin contained in the
preparation of the present invention may vary according to
various parameters, such as the nature of the preparation,

30

35

-4-

0115627

1 the particular kind or activity of calcitonin employed and
the condition or ailment to be treated with the preparations.
In general, the concentrations are somewhat higher than those
found in compositions for the systemic administration of
5 calcitonin. It has been found that a concentration level of
1 to 150 micrograms per ml and preferably 2 to 30 micrograms
per ml achieve the desired result. The levels of
administration of calcitonin also vary somewhat from those
used systemically. In the case of human patients, for
10 example, amounts of from 0.7 to 70 micrograms, particularly
from 1 to 25 micrograms, are usually appropriate for single
dosages given and repeated as often as the physician finds it
necessary and such dosages correspond generally to about 0.01
to 1 micrograms, and particularly 0.03 to 0.35 micrograms,
15 per kilogram of body weight. (The above concentration and
dosage levels of calcitonin apply to calcitonin with a
potency of about 4000 International Units per mg and may be
adjusted pro rata for calcitonin of other potencies.)

The diluent base or vehicle used in accordance with
20 the present invention may be non-aqueous or aqueous. In the
former case the group of diluents is the physiologically
acceptable polar solvents. Preferred compounds of this type
are those with which it is possible to make a solution of
adequate concentration of dissolved calcitonin. Examples of
25 these compounds include dimethylsulphoxide, dimethyl
formamide, dimethylauramide, polyhydroxy alcohols, vegetable
and mineral oils. If desired, such non-aqueous media may be
mixed with water to form the diluent of the preparation.

30

35

-5-

0115627

1 However, the degree of physiological acceptability of the non-aqueous diluents is generally less than that of aqueous media and the preferred diluent is therefore water without the addition of organic solvents.

5 In the preparations of the present invention, calcitonin is used in combination with an absorption promoter. Such absorption promoters include the physiologically acceptable surface active agents. The amount of such an agent may be in the range from about 0.01 to about 10% w/v or higher and preferably about 0.05 to about 1.0% w/v, the amount depending on the specific surfactant used. The amount is generally kept as low as possible since above a certain level no further enhancement of absorption can be achieved and also too high of a surfactant level may cause irritation of the nasal mucosa. Such surface active agents include:

- a. Bile salts, such as sodium taurocholate, sodium cholate, sodium deoxycholate and sodium glycholate;
- b. Cationics, such as the long chain amine condensates with ethylene oxide and quaternary ammonium compounds, for example cetyl trimethyl ammonium bromide and dodecyl dimethyl ammonium bromide;
- c. Anionics, such as alkylbenzenesulfonates, N-acyl-N-alkyltaurates, α -olefin sulfonates, sulfated linear primary alcohols and sulfated polyoxyethylenated straight-chain alcohols;
- d. Nonionics, such as polyoxyethylenated alkylphenols, polyoxyethylenated straight chain alcohols, long chain carboxylic acid esters including glycerol ester of natural fatty acids, propylene glycol, sorbitol, and polyoxyethylenated sorbitol esters;

-6-

0115627

- 1 e. Amphoterics, such as imidazoline carboxylates,
sulfonates and the like; and
f. Phospholipids, such as phosphotidyl choline and the like.

5 The preparations of the present invention preferably contain a phosphate or acetate buffer in the range of 0.01 M to 0.5 M and preferably in the range of 0.05 M to 0.2 M. This concentration was found effective to provide stability of the dissolved calcitonin in the diluent base or vehicle.

10 The preparations of the present invention may also contain other additives, such antioxidants, stabilizers, tonicity adjusters, viscosity builders, preservatives, and the like. The concentration of these additives may vary according to the particular additive used and the desired result sought. In general, the concentrations for these
15 additives will be in the range as follows:

<u>Additives</u>	<u>% W/V</u>
Antioxidants	0.01 - 0.2
Stabilizers	0.01 - 2.0
20 Tonicity Adjuster	0.01 - 0.5
Viscosity Builders	0.1 - 2.0
Preservatives	0.001 - 2.0

25 While the use of the kind and concentration of additives will be well within the ability of the skilled artisan, the following will serve as illustration for two additives generally used in pharmaceutical preparations intended for similar purposes.

30

35

-7-

0115627

1	<u>Preservatives</u>	<u>% W/V</u>
	Benzalkonium chloride	0.004 - 0.02
	Disodium Ethylene	
	Diamine Tetraacetate	0.01 - 0.2
5	Thimerosal	0.001 - 0.01
	Chlorobutanol	0.5 - 1.0
	Methyl and/or Propyl	
	Paraben	0.01 - 0.2
	Phenethyl Alcohol	0.25 - 0.75
10	Cyclohexedine	0.01 - 0.1
	<u>Viscosity Agents</u>	<u>% W/V</u>
	Methyl Cellulose	0.1 - 2.0
	Hydroxyethyl Cellulose	0.1 - 2.0
15	Hydroxypropyl Cellulose	0.1 - 2.0
	Polyvinylpyrrolidone	0.5 - 2.0

20 In preparing the formulations of the present invention, calcitonin is dissolved in the vehicle or diluent after which the additional ingredients are added in accordance with customary formulation procedures known in the pharmaceutical industry.

25 Examples of typical intranasal formulations are set forth below. However, it is to be understood that these examples are given by way of illustration only and are not to be construed as limiting the invention either in spirit or in scope as many modifications will be apparent to those skilled in the art.

30

35

-8-

0115627

1	<u>EXAMPLE 1</u>	<u>% W/V</u>
	Calcitonin	0.009
5	Sodium Taurocholate	0.5
	Gelatin	1.0
	Purified Water Q.S.	100
10	<u>EXAMPLE 2</u>	<u>% W/V</u>
	Calcitonin	0.009
	Miranol C2M	1.0
	Gelatin	1.0
15	Purified Water Q.S.	100
	<u>EXAMPLE 3</u>	<u>% W/V</u>
	Calcitonin	0.009
20	Miranol C2M	0.05
	Sodium Acetate .3H ₂ O	1.36
	Acetic Acid	0.6
	Purified Water Q.S.	100
25	<u>EXAMPLE 4</u>	<u>% W/V</u>
	Calcitonin	0.009
	Polysorbate 80	1.0
30	Sodium Acetate .3H ₂ O	1.36
	Acetic Acid	0.6
	Purified Water Q.S.	100

35

-9-

0115627

1

EXAMPLE 5% W/V

5

Calcitonin	0.003
Brij 30	1.0
Sodium Acetate .3H ₂ O	1.36
Acetic Acid	0.6
Purified Water Q.S.	100

10

EXAMPLE 6% W/V

15

Calcitonin	0.009
Myrj 59	1.0
Sodium Acetate	1.36
Acetic Acid	0.6
Purified Water Q.S.	100

20

EXAMPLE 7% W/V

25

Calcitonin	0.009
Miranol C2M	1.0
Sodium Phosphate	2.40
Citric Acid	0.34
Thimerasol	0.002
Purified Water Q.W.	100

30

35

-10-

0115627

1	<u>EXAMPLE 8</u>	<u>% W/V</u>
	Calcitonin	0.009
5	Sodium Taurocholate	0.5
	Sodium Acetate .3H ₂ O	1.36
	Acetic Acid	0.6
	Benzalkonium Chloride	0.01
	DiSodium ethylenediame	
10	tetraacetate	0.1
	Purified Water Q.S.	100
	<u>EXAMPLE 9</u>	<u>% W/V</u>
15	Calcitonin	0.009
	Sodium Taurocholate	0.5
	Sodium Acetate .3H ₂ O	1.36
	Acetic Acid	1.36
	Chlorobutanol	0.1
20	Phenethyl Alcohol	0.2
	Purified Water Q.S.	100
	<u>EXAMPLE 10</u>	<u>% W/V</u>
25	Calcitonin	0.003
	Miranol C2M	1.0
	Sodium Phosphate	2.40
	Citric Acid	0.34
	Thimerasol	0.002
30	Purified Water Q.S.	100

35

-11-

0115625

- 1 The gelatin used in the above formulations is a
standard hydrolipid animal gelatin prepared for
pharmaceutical use and routinely used as a diluent for
peptides.
- 5 According to the present invention, it has been
found that calcitonin can be administered intranasally from a
vehicle containing absorption promoters with results
considerably superior to those obtained with the
administration of calcitonin without absorption promoters.
- 10 The following studies were undertaken to examine the
bioavailability of calcitonin from the formulations of the
presetn invention, dependency of intranasal absorption of
calcitonin on the level of absorption promoters and stability
of calcitonin in the presence of absorption promoters.
- 15 PROTOCOL
- Male rats weighing 150-250 g were weighed and
anesthetized with sodium pentobarbital, 50/mg/kg. by
intraperitoneal injection. Once anesthetized the
nasopalatine process was occluded with glue. The animals
20 were randomly placed into groups of 5-7 rats with the number
of groups being dependent upon the number of intranasal
formulations to be tested. Supplemental pentobarbital
anesthesia was administered as necessary throughout the
study.
- 25 Prior to administration of the test material, blood
was collected by cardiac puncture using a 25G 5/8" needle.
Fifty (50) microliters of the salmon calcitonin-containing
surfactant solution was then instilled into the nasal septum
using polyethylene tubing (PE 20, Peterson Technics, Monmouth
30 Junction, N.J.) connected to a 1 ml syringe; the tubing was
inserted about 1 cm into the nasal septum. One and three

-12-

0115627

- 1 hours after nasal instillation, blood was again collected by
cardiac puncture.

Biochemical Analysis

- 5 Blood samples were allowed to clot at room
temperature and were then refrigerated for 30-60 minutes to
provide maximum clot retraction. The samples were
centrifuged at 4°C., 5000 rpm for 10 minutes (Beckman Model
J2-21 Centrifuge, Beckman Instruments, Palo Alto, CA). Serum
calcium was quantitated using a Calcette (Model 4008,
10 Precision Systems, Sudbury, MA).

Data Analysis

- 15 Serum calcium values at 0, 1 and 3 hours were
expressed as mean \pm standard deviation. In addition, the
absolute change and the percent change from the pretreatment
(0 time) value at 1 and 3 hours was also calculated.
Statistical analysis consisted of comparison of the serum
calcium values at 0 and 1 hour, 0 and 3 hours, and 1 and 3
hours using a t test.

20

25

30

35

-13-

0115627

1

EXAMPLE 11

This example illustrates decrease in serum calcium in blood samples obtained in accordance with the above protocol when: a. calcitonin is administered alone;

5

b. calcitonin is administered in formulations containing various absorption promoters; and c. no calcitonin is present in the formulations.

Table 1 shows the result obtained.

10

15

20

25

30

35

-14-

0115627

TABLE I

Calcitonin U/kg body weight vehicle/surfactant	TIME AFTER DOSE			
	0 hour mg/dl	1 hour mg/dl	% dect.	3 hour mg/dl % decrease
2U 1% gel -	8.8	9.5	NONE	9.9 NONE
5U 1% gel -	8.5	7.9	7.1	9.5 NONE
10U 1% gel -	9.2	7.6	17.4	9.7 NONE
10U .1M Acetate -	8.8	6.3	28.4	9.0 NONE
- 1% gel 1% Miranol G2M(1)	8.9	8.0	NONE	9.2 NONE
- 1% gel 1% Taurocholate	9.1	9.5	NONE	9.8 NONE
3U 1% gel 1% Miranol G2M(1)	8.9	6.7	24.7	7.6 14.6
	8.8	6.5	26.1	8.5 3.4
	8.7	6.8	21.8	8.9 2.3
3U 0.1M Acet. 1% Miranol G2M(1)	8.7	6.8	21.8	8.9 2.3
10U 1% gel 1% Miranol G2M(1)	9.5	6.1	35.8	8.8 7.4
	9.1	7.5	17.6	6.6 27.5
	8.9	7.1	20.2	7.1 20.2
10U 0.1M Acet. 1% Miranol G2M(1)	9.3	6.1	34.4	6.5 30.1
	9.2	6.8	26.1	8.4 8.7
3U 1% gel 1% Taurocholate	9.1	7.0	23.1	6.4 29.6
10U 1% gel 1% Taurocholate	9.3	7.1	23.7	6.3 32.3
	8.6	6.1	29.1	5.9 31.4
10U 0.1M Acet. 1% Taurocholate	9.1	6.5	28.6	5.7 37.4
3U 1% gel 1% Tween 80 (Polysorbate 80)	8.3	6.2	25.3	8.7 NONE
	8.9	7.1	20.2	9.2 NONE

(1) dicarboxylated fatty imidazoline or dicarboxylic coconut derivative,

-15-

0115627

TABLE I (Cont'd.)

Calcitonin U/kg body weight vehicle/surfactant	0 hour mg/dl	TIME AFTER DOSE			% decrease
		1 hour mg/dl	% decr.	3 hour mg/dl	
100 1% gel 1% Tween 80 (Polysorbate)	8.7	6.6	24.2	7.3	16.1
30 1% gel 0.5% Benzal- konium Chloride	8.9	6.7	24.7	8.4	5.6
	8.7	5.9	32.2	6.3	27.6
100 1% gel 0.5% Benzal- konium Chloride	8.9	6.5	26.9	9.0	NONE
	8.5	7.2	15.3	7.9	7.1
30 1% gel 1% Saponin (Sapogen Glycoside)	9.0	6.0	33.3	6.2	31.0
100 .1M Acet. 1% NaI Saf	8.6	7.3	15.1	7.3	15.1
100 .1M Acet. 1% Br13 30	8.7	6.5	25.9	8.1	6.9
(Polyoxyethylene (4) lauryl ether)	8.7	6.5	25.9	6.5	25.9
100 .1M Acet. 1% Myrj 59	8.5	6.2	27.1	8.4	1.2
(Polyoxyethylene (100) Stearate)					
100 .1M Acet. 1% Tween 80	8.7	7.5	13.8	7.1	18.4
100 .1M Acet. 1% Aer OT	9.1	6.6	27.5	7.6	16.5
(Sodium dioctyl sulfosuccinate)					

-16-

0115627

1

EXAMPLE 12

This example illustrates that the enhancement of intranasal absorption depends on the level of absorption promoter present in the formulation.

5

Table II shows the result obtained.

10

15

20

25

30

35

-17-

0115627

1

5

10

15

20

25

30

35

TABLE IX

10U Calcitonin/kilo in 0.1M Acetate with	0 hour mg/dl	1 hour mg/dl	3 hour mg/dl	%
1% Taurocholate	9.1	6.5	5.7	37.4
0.5%	9.0	6.1	7.5	16.6
0.25%	9.1	6.8	7.4	18.2
0.1%	8.9	6.5	8.3	6.7
0.05%	9.0	7.6	8.7	3.3

10U calcitonin/kilo
in 0.1M Acetate with

1% Miranol C2H (dicarboxylic coconut derivative, sodium salt)

0.5% "

0.25% "

0.1% "

0.5% "

-18-

0115627

1

EXAMPLE 13

This example illustrates that calcitonin maintains its activity level in the formulations of the present invention on storage at room temperatures.

5

Table III shows the results obtained.

10

15

20

25

30

35

-19-

0115527

TABLE III

10U calcitonin in 1% gel with 1% Mirunol C2M		0 hour mg/dl	1 hour mg/dl	3 hour mg/dl	%
Initial		8.9	7.1	7.1	20.2
2 wks @ RT		9.1	7.2	6.4	29.7
4 wks @ RT		9.1	6.0	6.9	24.2
10U calcitonin in 1% gel with 1% Tween 80 (Polysorbate 80)					
Initial		8.9	6.7	8.4	5.6
2 wks @ RT		8.8	6.7	8.3	6.0
4 wks @ RT		8.8	6.3	6.7	23.9

-20-

0115627

1 What is claimed is:

1. A pharmaceutical composition for the treatment of disorders of bone metabolism which comprises an aqueous or non-aqueous medium suitable for intranasal administration and containing a therapeutically effective amount of calcitonin and a surface active agent.

2. The pharmaceutical composition of claim 1 further comprising a buffer.

3. The pharmaceutical composition of claim 2 wherein said buffer is from 0.01 to 0.5M.

4. The pharmaceutical composition of any of claims 1-3 further comprising an antioxidant, stabilizer, tonicity adjuster, viscosity builder, or a preservative.

5. The pharmaceutical composition of any of claims 1-4 wherein said medium contains from about 5 to about 150 micrograms calcitonin per ml of said aqueous medium.

6. The pharmaceutical composition of any of claims 1-5 containing from about 0.01 to about 10% w/v of the surface active agent.

7. The pharmaceutical composition of claim 2 wherein said buffer is a phosphate buffer.

8. The pharmaceutical composition of claim 2 wherein said buffer is an acetate buffer.

9. The pharmaceutical composition of any of claims 1-8 wherein said aqueous medium is a gel.

10. The pharmaceutical composition of any of claims 1-9 wherein said surface active agent is a dicarboxylated fatty imidazoline or sodium taurocholate, or a benzalkonium chloride.

30

35

European Patent
Office

EUROPEAN SEARCH REPORT

0115627

Application number

DOCUMENTS CONSIDERED TO BE RELEVANT			EP 83113070.3
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int. Cl. 7)
A	<u>US - A - 4 241 051 (CHRISTIE et al.)</u> * Claims 3,4,5,7-10; column 1, line 45 - column 4, line 46 * --	1,6	A 61 K 37/02
A	<u>GB - A - 1 548 984 (CIBA-GEIGY AG)</u> * Claims, especially claim 5; page 1, line 9 - page 3, line 66 * --	1,2,5	
A	<u>DE - A1 - 2 254 061 (HOECHST AG)</u> * Claim 1,4; pages 1,2 * ----		
			TECHNICAL FIELDS SEARCHED (Int. Cl. 7)
			A 61 K 37/00
The present search report has been drawn up for all claims			
Place of search VIENNA		Date of completion of the search 30-03-1984	Examiner STÖCKLMAYER
CATEGORY OF CITED DOCUMENTS X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure F : intermediate document T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons & : member of the same patent family, corresponding document			